

AMENDMENTS TO THE CLAIMS:

The following listing of claims replaces all prior versions of the claims.

LISTING OF CLAIMS:

1-88. (canceled).

89. (not entered) A method for preparing a porcine embryo capable of developing into a live-born porcine animal comprising:

(a) culturing a diploid differentiated porcine nuclear donor cell;

(b) inserting the nucleus of the cultured diploid differentiated porcine nuclear donor cell into an unactivated, enucleated metaphase II-arrested porcine oocyte to reconstruct an embryo,

wherein the nuclear donor cell is in the G1 phase of the cell cycle; and

(c) activating the reconstructed embryo to establish the porcine embryo.

90. (not entered) The method according to claim 89, wherein the cell is cultured in TC medium 199.

91. (not entered) The method according to claim 89, wherein the cultured diploid differentiated porcine nuclear donor cell is a fibroblast cell.

92. (not entered) The method according to claim 89, wherein the cultured diploid differentiated porcine nuclear donor cell is a transgenic cell.

93. (not entered) The method according to claim 89, wherein the enucleated porcine oocyte is prepared by maturing the oocyte for 48 hours and enucleating the oocyte.

94. (not entered) The method according to claim 89, wherein step (b) comprises electrically fusing the cell and the enucleated oocyte.

95. (not entered) The method according to claim 89, wherein the embryo is transferred into an oviduct of a recipient porcine female.

96. (not entered) The method according to claim 89, wherein the embryo is transferred into a uterine horn of a recipient porcine female.

97. (not entered) The method according to claim 96, wherein the embryo is a blastocyst.

98. (not entered) A method of cloning a pig by nuclear transfer comprising:

(a) inserting a nucleus of a cultured diploid porcine differentiated cell in the G1 phase of the cell cycle into an unactivated, enucleated metaphase II-arrested porcine oocyte to reconstruct an embryo;

(b) activating the resultant reconstructed embryo;

(c) transferring the activated, reconstructed embryo to a host pig such that the reconstructed embryo develops to term.

99. (not entered) The method according to claim 98, wherein the cell is cultured in TC medium 199.

100. (not entered) The method according to claim 98, wherein the cultured diploid differentiated porcine nuclear donor cell is a fibroblast cell.

101. (not entered) The method according to claim 98, wherein the cultured diploid differentiated porcine nuclear donor cell is a transgenic cell.

102. (not entered) The method according to claim 98, wherein the enucleated porcine oocyte is prepared by maturing the oocyte for 48 hours and enucleating the oocyte.

103. (not entered) The method according to claim 98, wherein step (a)

comprises electrically fusing the cell and the enucleated oocyte.

104. (not entered) The method according to claim 98, wherein the cultured, reconstructed embryo is transferred into a uterine horn of a recipient porcine female.

105. (not entered) A method for preparing a porcine embryo, comprising:

(a) culturing one or more nonembryonic porcine cells in a medium comprising one or more components selected from the group consisting of LIF, FGF and stem cell factor to obtain one or more cultured cells;

(b) translocating one of said cultured cells, or a nucleus thereof, into an enucleated porcine oocyte to establish a nuclear transfer oocyte; and

(c) activating said nuclear transfer oocyte to establish said porcine embryo.

106. (not entered) A method according to claim 105, wherein said cell culture medium further comprises between 10 mM and 100 mM glucose.

107. (not entered) A method according to claim 106, wherein said cell culture medium comprises 25 mM glucose.

108. (not entered) A method according to claim 105, wherein said cell culture medium comprises between 10 and 100 ng/mL of each of said LIF, FGF, and stem cell factor.

109. (not entered) A method according to claim 108, wherein said medium comprises 20 ng/mL of each of said LIF, FGF, and stem cell factor.

110. (not entered) A method according to claim 105, wherein said one or more nonembryonic porcine cells are one or more fetal cells.

111. (not entered) A method according to claim 110, wherein said one or more porcine fetal cells are obtained from the genital ridge.

112. (not entered) A method according to claim 105, wherein said cultured cell in step (b) is a transgenic cell.

113. (not entered) A method for preparing a cloned porcine embryo, comprising:
(a) translocating a cultured nonembryonic porcine cell, or a nucleus thereof, into an enucleated porcine oocyte to establish a nuclear transfer oocyte, wherein said porcine oocyte is a sow oocyte; and

(b) activating said nuclear transfer oocyte to establish said porcine embryo.

114. (not entered) A method according to claim 113, wherein said one or more nonembryonic porcine cells are one or more fetal cells.

115. (not entered) A method according to claim 114, wherein said one or more porcine fetal cells are obtained from the genital ridge.

116. (not entered) A method according to claim 113, wherein said cultured nonembryonic porcine cell in step (a) is a transgenic cell.

117. (not entered) A method according to claim 113, wherein said enucleated porcine oocyte is prepared by the method comprising:

(a) maturing said sow oocyte for between 41 and 54 hours; and

(b) enucleating said sow oocyte.

118. (not entered) A method according to claim 105 or 113 wherein said translocation step comprises:

placing said cell within the perivitelline space of said enucleated oocyte; and electrically fusing said cell and said enucleated oocyte.

119. (not entered) A method for preparing a porcine embryo, comprising:

(a) culturing one or more nonembryonic porcine cells in a medium comprising between 10 mM and 100 mM glucose;

(b) translocating one of said cultured cells, or a nucleus thereof, into an enucleated porcine oocyte to establish a nuclear transfer oocyte; and

(c) activating said nuclear transfer oocyte to establish said porcine embryo.

120. (not entered) A method according to claim 119, wherein said cell culture medium comprises 25 mM glucose.

121. (not entered) A method according to claim 119, wherein said one or more nonembryonic porcine cells are one or more fetal cells.

122. (not entered) A method according to claim 121, wherein said one or more porcine fetal cells are obtained from the genital ridge.

123. (not entered) A method according to claim 119, wherein said cultured nonembryonic porcine cell in step (a) is a transgenic cell.

124. (not entered) A method according to claim 105, 113, or 119, wherein said activation step comprises:

(a) incubating said nuclear transfer oocyte in a medium comprising between 10 and 20 μ M ionomycin; and

(b) incubating said nuclear transfer oocyte in a medium comprising between 1 and 4 mM DMAP.

125. (not entered) A method for preparing a porcine fetus, comprising:
transferring a porcine embryo produced according to any one of claims 105, 113, or 119 into a recipient porcine female to produce said porcine fetus, wherein said porcine embryo and said recipient porcine female are asynchronous.

126. (not entered) A method according to claim 125, wherein said porcine embryo is one of a plurality of porcine embryos transferred into said recipient porcine female.

127. (not entered) A method according to claim 126, wherein 90 or more embryos are transferred into said recipient porcine female.

128. (not entered) A method according to claim 125, wherein said embryo is transferred into an oviduct said recipient porcine female.

129. (not entered) A method according to claim 128, wherein said embryo comprises from 1 to 3 cells.

130. (not entered) A method according to claim 126, wherein said embryo is transferred into a uterine horn of said recipient porcine female.

131. (not entered) A method according to claim 130, wherein said embryo comprises 3 or more cells.

132. (not entered) A method for preparing a porcine animal, comprising:

- a) translocating a cultured nonembryonic porcine cell[s], or a nucleus thereof, into an enucleated porcine oocyte to establish a nuclear transfer oocyte;
- b) activating said nuclear transfer oocyte to establish said porcine embryo; and
- c) transferring said porcine embryo into a recipient female to produce said porcine animal, wherein said porcine embryo and said recipient female are asynchronous.

133. (not entered) A method according to claim 132, wherein said cultured nonembryonic porcine cell is cultured in a medium comprising one or more components selected from the group consisting of LIF, FGF and stem cell factor.

134. (not entered) A method according to claim 132, wherein said enucleated porcine oocyte is prepared by the method comprising:

- (a) obtaining an oocyte from a sow;
- (b) maturing said oocyte for between 41 and 54 hours; and
- (c) enucleating said oocyte.

135. (not entered) A method according to claim 132, wherein said translocation step comprises:

placing said cell, or a nucleus thereof, within the perivitelline space of said enucleated oocyte; and

electrically fusing said cell and said enucleated oocyte.

136. (not entered) A method according to claim 132, wherein said activation step comprises:

(a) incubating said nuclear transfer oocyte in a medium comprising between 10 and 20 μ M ionomycin; and

(b) incubating said nuclear transfer oocyte in a medium comprising between 1 and 4 mM DMAP.

137. (not entered) A method according to claim 132, wherein said cell culture medium further comprises between 10 mM and 100 mM glucose.

138. (not entered) A method according to claim 137, wherein said cell culture medium comprises about 25 mM glucose.

139. (not entered) A method according to claim 132, wherein said cell culture medium comprises between 10 and 100 ng/mL of each of said LIF, FGF, and stem cell factor.

140. (not entered) A method according to claim 139, wherein said medium comprises about 20 ng/mL of each of said LIF, FGF, and stem cell factor.

141. (not entered) A method according to claim 132, wherein said one or more nonembryonic porcine cells are porcine fetal cells.

142. (not entered) A method according to claim 132, wherein said porcine fetal cells are obtained from the genital ridge.

143. (not entered) A method according to claim 132, wherein said porcine embryo is more developed than would be expected if said recipient porcine female and said porcine embryo were synchronous.

144. (not entered) A method according to claim 143, wherein said porcine embryo is activated between about 1 day prior to the onset of standing estrus in said recipient porcine female and about 43 hours after the onset of standing estrus in said recipient porcine female.

145. (not entered) A method according to claim 143, wherein said porcine embryo is activated between about 18 hours prior to the onset of standing estrus in said recipient porcine female and about 24 hours after the onset of standing estrus in said recipient porcine female.

146. (not entered) A method according to claim 132, wherein said porcine embryo is one of a plurality of porcine embryos transferred into said recipient porcine female.

147. (not entered) A method according to claim 146, wherein 90 or more embryos are transferred into said recipient porcine female.

148. (not entered) A method according to claim 132, wherein said embryo is transferred into an oviduct of said recipient porcine female.

149. (not entered) A method according to claim 148, wherein said embryo comprises from 1 to 3 cells.

150. (not entered) A method according to claim 132, wherein said embryo is transferred into a uterine horn of said recipient porcine female.

151. (not entered) A method according to claim 150, wherein said embryo comprises 3 or more cells.

152. (new) A live-born clone of a pre-existing, non-embryonic, donor mammal from which a differentiated cell has been taken,

wherein the mammal is selected from cattle, sheep, pigs, goats, mice, rabbits, horses, and rats,

wherein the clone is produced by a process comprising:

(a) transferring the nucleus of the differentiated cell or a cell obtained by culture thereof into an enucleated, metaphase II-arrested oocyte from the same species,

wherein the differentiated cell or cell obtained by culture thereof is a diploid cell in the G1 phase of the cell cycle at the time of transfer;

(b) activating the oocyte; and

(c) incubating the activated oocyte such that an embryo develops;

(d) transferring the embryo to a female of the same species; and

(e) developing the embryo into the live-born clone.

153. (new) The clone of claim 152, wherein the differentiated cell or cell obtained by culture thereof is cultured *in vitro*.

154. (new) The clone of claim 152, wherein the differentiated cell or cell obtained by culture thereof is abstracted *ex vivo*.

155. (new) The clone of claim 152, wherein the mammal is a cattle.

156. (new) The clone of claim 152, wherein mammal is a sheep.

157. (new) The clone of claim 152, wherein the mammal is a pig.

158. (new) The clone of claim 152, wherein the mammal is a goat.

159. (new) The clone of claim 152, wherein the mammal is a mouse.

160. (new) The clone of claim 152, wherein the mammal is a rabbit.

161. (new) The clone of claim 152, wherein the mammal is a horse.

162. (new) The clone of claim 152, wherein the mammal is a rat.

163. (new) A live-born clone of a pre-existing, non-embryonic, donor mammal, wherein the mammal is selected from cattle, sheep, pigs, goats, mice, and rabbits, horses, and rats.

164. (new) The clone of claim 163, wherein the mammal is a cattle.

165. (new) The clone of claim 163, wherein mammal is a sheep.

166. (new) The clone of claim 163, wherein the mammal is a pig.

167. (new) The clone of claim 163, wherein the mammal is a goat.

168. (new) The clone of claim 163, wherein the mammal is a mouse.

169. (new) The clone of claim 163, wherein the mammal is a rabbit.

170. (new) The clone of claim 163, wherein the mammal is a horse.

171. (new) The clone of claim 163, wherein the mammal is a rat.